JC14 Rec'd PCT/PTO 18 DEC 2001

FORM PTO-1390 (REV 11-2000) U S DEPARTMENT OF COMMERCE PATENT AND TRADEMARK OFFICE

TRANSMITTAL LETTER TO THE UNITED STATES DESIGNATED/ELECTED OFFICE (DO/EO/US) CONCERNING A FILING UNDER 35 U.S.C. § 371

ATTORNEY'S DOCKET NUMBER: 350292001100

U S APPLICATION NO (If known, see 37 CFR 1 5): 10/018293 (to be assigned)

INTERNATIONAL APPLICATION NO.	INTERNATIONAL FILING DATE	PRIORITY DATE CLAIMED
PCT/JP00/04022	June 20, 2000	June 22, 1999

TITLE OF INVENTION: METHOD OF GENERATING VIRUS-FREE PLANTS

APPLICANT(S) FOR DO/EO/US: Masanori AYABE et al.

Applicant herewith submits to the United States Designated/Elected Office (DO/EO/US) the following items and other information

1. ☒ This is a **FIRST** submission of items concerning a filing under 35 U.S C 371
2. ☐ This is a **SECOND** or **SUBSEQUENT** submission of items concerning a filing under 35 U.S.C. 371.
3. ☒ This is an express request to begin national examination procedures (35 U.S.C. 371(f)). The submission must include items (5), (6), (9) and (21) indicated below.
4. ☐ The US has been elected by the expiration of 19 months from the priority date (PCT Article 31)
5. ☒ A copy of the International Application as filed (35 U.S.C. 371(c)(2))
   - a. ☐ is attached hereto (required only if not communicated by the International Bureau).
   - b. ☒ has been communicated by the International Bureau.
   - c. ☐ is not required, as the application was filed in the United States Receiving Office (RO/US)
6. ☒ An English language translation of the International Application under PCT Article 19 (35 U.S.C. 371(c)(2)).
   - a. ☒ is attached hereto.
   - b. ☐ has been previously submitted under 35 U.S.C. 154(d)(4).
7. ☒ Amendments to the claims of the International Application under PCT Article 19 (35 U.S.C. 371(c)(3)).
   - a. ☐ are attached hereto (required only if not communicated by the International Bureau).
   - b. ☐ have been communicated by the International Bureau.
   - c. ☐ have not been made; however, the time limit for making such amendments has NOT expired.
   - d. ☒ have not been made and will not be made.
8. ☐ An English language translation of the amendments to the claims under PCT Article 19 (35 U.S.C 371(c)(3)).
9. ☒ An oath or declaration of the inventor(s) (35 U.S.C. 371(c)(4)).
10. ☒ An English language translation of the annexes to the International Preliminary Examination Report under PCT Article 36 (35 U.S.C. 371(c)(5)).

Items 11. to 16. below concern document(s) or information included:

11. ☒ An Information Disclosure Statement under 37 CFR 1.97 and 1.98.
12. ☒ An assignment document for recording. A separate cover sheet in compliance with 37 CFR 3.28 and 3.31 is included.
13. ☐ A FIRST preliminary amendment.
14. ☐ A SECOND or SUBSEQUENT preliminary amendment.
15. ☐ A substitute specification.
16. ☐ A change of power of attorney and/or address letter.
17. ☒ A computer-readable form of the sequence listing in accordance with PCT Rule 13ter.2 and 35 U.S.C. 1.821 - 1.825
18. ☐ A second copy of the published international application under 35 U.S.C. 154(d)(4).
19. ☐ A second copy of the English language translation of the international application under 35 U.S.C. 154(d)(4).
20. ☒ Other items or information: a copy of the front page of published PCT application; international search report; Form PCT/IB/308; and return receipt postcard.

CERTIFICATE OF HAND DELIVERY

I hereby certify that this correspondence is being hand filed with the United States Patent and Trademark Office in Washington, D.C. on December 18, 2001.

Jinrong Li

dc-292355

JC05 Rec'd PCT/PTO 2001

PLICATION NO. (if known, see 37 CFR 1.5) to be Assigned 10/018293	INTERNATIONAL APPLICATION NO. PCT/JP00/04022	ATTORNEY'S DOCKET NUMBER: 350292001100

21. ☐ The following fees are submitted:

BASIC NATIONAL FEE (37 CFR 1.492(a)(1)-(5)):

Neither international preliminary examination fee (37 CFR 1.482) nor international search fee (37 CFR 1.445(a)(2)) paid to USPTO and International Search Report not prepared by the EPO or JPO.....$1,040.00

International preliminary examination fee (37 CFR 1.482) not paid to USPTO but International Search Report prepared by the EPO or JPO.....$890.00

International preliminary examination fee (37 CFR 1.482) not paid to USPTO but international search fee (37 CFR 1.445(a)(2)) paid to USPTO.....$740.00

International preliminary examination fee (37 CFR 1.482) paid to USPTO but all claims did not satisfy provision of PCT Article 33(1)-(4).....$710.00

International preliminary examination fee (37 CFR 1.482) paid to USPTO and all claims satisfied provisions of PCT Article 33(1)-(4).....$100.00

	CALCULATIONS PTO USE ONLY	
ENTER APPROPRIATE BASIC FEE AMOUNT =	$890.00	
☐ Surcharge of **$130.00** for furnishing the oath or declaration later than ☐ 20 ☐ 30 months from the earliest claimed priority date (37 CFR 1.492(e)).	$*	

CLAIMS	NUMBER FILED	NUMBER EXTRA	RATE		
Total claims	8 - 20 =	*	x $18.00	$*	
Independent claims	1 - 3 =	*	x $84.00	$*	
MULTIPLE DEPENDENT CLAIM(S) (if applicable)			+ $280.00	$280.00	
TOTAL OF ABOVE CALCULATIONS =				$280.00	
☐ Applicant claims small entity status. See 37 CFR 1.27. The fees indicated above are reduced by ½.				$*	
SUBTOTAL =				$	
Processing fee of **$130.00** for furnishing the English translation later than ☐ 20 ☐ 30 months from the earliest claimed priority date (37 CFR 1.492(f)). +				$*	
TOTAL NATIONAL FEE =				$1,170.00	
Fee for recording the enclosed assignment (37 CFR 1.21(h)). The assignment must be accompanied by an appropriate cover sheet (37 CFR 3.28, 3.31). **$40.00 per property** +				$40.00	
TOTAL FEES ENCLOSED =				$1,210.00	
				Amount to be refunded:	$*
				charged:	$*

a. ☐ A check in the amount of $* to cover the above fees is enclosed.

b. ☒ Please charge my **Deposit Account No. 03-1952** in the amount of $1,210.00 to cover the above fees. A duplicate copy of this sheet is enclosed.

c. ☒ The Commissioner is hereby authorized to charge any additional fees that may be required, or credit any overpayment to **Deposit Account No. 03-1952** referencing Docket No. 350292001100. A duplicate copy of this sheet is enclosed.

d. ☐ Fees are to be charged to a credit card. **WARNING:** Information on this form may become public. Credit card information should not be included on this form. Provide credit card information and authorization on PTO-2038.

NOTE: Where an appropriate time limit under 37 CFR 1.494 or 1.495 has not been met, a petition to revive (37 CFR 1.137(a) or (b)) must be filed and granted to restore the application to pending status.

SEND ALL CORRESPONDENCE TO:

Barry E. Bretschneider
Morrison & Foerster LLP
2000 Pennsylvania Avenue, N.W.
Washington, D.C. 20006-1888

SIGNATURE

Barry E. Bretschneider
Registration No. 28,055

dc-292355

10/018293
JC05 Rec'd PCT/PTO 6 DEC 2001

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Atty. Docket No: 35029-20011.00

In re patent application of

AYABE, MASANORI *et al.*

Serial No. Unassigned

Filed: Concurrently Herewith

For: MEHTHOD OF GENERATING VIRUS-FREE PLANTS

STATEMENT TO SUPPORT FILING AND SUBMISSION IN ACCORDANCE WITH 37 C.F.R. §§ 1.821-1.825

Assistant Commissioner for Patents
Washington, D.C. 20231
Box SEQUENCE

Sir:

In connection with a Sequence Listing submitted concurrently herewith, the undersigned hereby states that:

1. the submission, filed herewith in accordance with 37 C.F.R. § 1.821(g), does not include new matter;

2. the content of the attached paper copy and the attached computer readable copy of the Sequence Listing, submitted in accordance with 37 C.F.R. § 1.821(c) and (e), respectively, are the same; and

3. all statements made herein of their own knowledge are true and that all statements made on information and belief are believed to be true; and further, that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United

States Code and that such willful false statements may jeopardize the validity of the application or any patent resulting therefrom.

Respectfully submitted,

Dec. 17, 2001
Date

James A. Coburn

HARBOR CONSULTING
Intellectual Property Services
1500A Lafayette Road
Suite 262
Portsmouth, N.H.
800-318-3021

PCT10

RAW SEQUENCE LISTING DATE: 01/09/2002
PATENT APPLICATION: **US/10/018,293** TIME: 11:53:41

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  4       SUMI, SHINICHIRO
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L:10 M:270 C: Current Application Number differs, Replaced Application Number
L:11 M:271 C: Current Filing Date differs, Replaced Current Filing Date

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10/018293
JC05 Rec'd PCT/PTO 18 DEC 2001

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SUMI, SHINICHIRO

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Statement

To: Director-General of the Patent Office Takahiko Kondo

This is to declare that the base sequences and the amino acid sequences stored in this flexible disk attached are the faithful codings of the base sequences and the amino acid sequences described in the specification and that no modifications to them have been made.

June 20, 2000

Indication of International Application:
International application filed on June 20, 2000
Docket No. H740-PCT
Title of the Invention: Method of generating virus-free plants
Applicant: Wakunaga Pharmaceutical Co., Ltd.
Attorney: Takashi Ishida

Rec'd PCT/PTO 26 JUN 2000

Document Describing the Information on the Storage Mode of the Flexible Disk and the Like

1. Name of Applicant: Wakunaga Pharmaceutical Co., Ltd.
2. Name of Attorney: Takashi Ishida
3. Indication of International Application:
   International application filed on June 20, 2000
   Docket No. H740-PCT
4. Title of the Invention: Method of generating virus-free plants
5. Letter Code Used: Shift JIS Code
6. Name of File Storing the Sequences: H740-PCT.TXT
7. Correspondence:
   Phone Number: 03(5470)1900
   Person in Charge: Tsumoru Fukumoto

PCT/JP00/03961

DESCRIPTION

METHOD OF GENERATING VIRUS-FREE PLANTS

Technical Field

The present invention relates to methods of generating virus-free plants of the genus *Allium* represented by *Allium sativum* and other plants that propagate via scaly bulbs and bulbs. More specifically, the present invention relates to methods of generating virus-free plants by isolating and culturing domy tissues obtained by culturing foliage leaf bases.

Background Art

Plants belonging to the genus *Allium* include garlic, onion, scallion, cibol, shallot, leek, chive and the like. They are widely used as edible plants and spice. Since ancient days garlic has been recognized to have medicinal values, and is also used as aphrodisiac, antasthenic, and the like. Plants of the genus *Allium* have been cultured in many parts of the world including Japan. In recent years, however, damages by viruses are causing serious problems in their culturing. As such viruses for *Allium sativum*, for example, there are known garlic viruses (GarVs), leek yellow stripe virus (LYSV), onion yellow dwarf virus (OYDV), garlic latent virus (GLV) and the like. It is known that garlic is a crop that grows through vegetative propagation and thus, once it is infected with a virus, the infection is inherited to later generations resulting in the spread of viral pollution. Similar damage is known for plants belonging to the genus *Allium* other than garlic, and there is an urgent need for essential measures to combat such viral pollutions.

On the other hand, tissue culture methods for plants were recently established, the application of which method allows elimination of viruses for various plants

that grow via vegetative propagation. It is generally known that even virus-infected plants have no viruses present in the shoot apex (meristem). Thus, by culturing the shoot apex and thereby redifferentiating the plants, plants that are not infected with virus (virus-free plants) can be obtained. It is also known that by culturing a plant tissue to form a callus and then subculturing the callus, virus can be eliminated. By redifferentiating the plant from the virus-free callus, virus-free plants can be obtained.

For plants of the genus *Allium* represented by *Allium sativum*, the above method has been employed to eliminate viruses, and in fact virus-free *Allium* plants have been cultivated.

However, by the method of culturing the shoot apex to obtain virus-free *Allium* plants, one shoot apex usually produces only one or a few plants. In order to obtain a multiplicity of plants, many shoot apexes must be extracted. Furthermore, since the shoot apex is located at the base of *Allium* ramentum and its size is about 0.5 mm or less, the extraction of the shoot apex requires an operation under the microscope. Thus, the operation of extracting shoot apexes is troublesome and the work efficiency is low.

On the other hand, in the method of redifferentiating the callus to obtain virus-free *Allium* plants, it is possible to grow the callus in large quantities once the callus has been induced. However, due to mutations frequently encountered during callus cultivation, it is difficult to obtain *Allium* plants of homogeneous genetic traits.

Accordingly, a tissue culture method for culturing in large quantities an *Allium* plant that was rendered virus-free by the shoot apex culture and a method of preparing redifferentiated plants using the base of foliage leaves were developed, which made possible to grow a large quantity of virus-free plants starting with

a small amount thereof as the material (Japanese Unexamined Patent Publication (Kokai) No. 6-197650). However, these methods require, at all times, virus-free plants that are used as the material, which in turn requires maintaining said plants in an isolated cultivation using a net house.

Disclosure of the Invention

The present invention relates to a culture method for generating virus-free plants from a tissue other than the shoot apex based on the plants infected with virus.

By culturing the base of foliage leaves, plant differentiation can be induced via a domy tissue. The inventors of the present invention have found for the first time that the domy tissue is undergoing active cell division and no virus is present therein, as in the shoot apex. Thus, by isolating only the domy tissue and culturing it, it has become possible to generate virus-free plants. Since a plurality of domy tissues are formed from one ramentum, the efficiency becomes dramatically enhanced as compared to the conventional method of shoot apex culture.

Thus, the present invention provides a method of generating virus-free plants characterized in that a domy tissue formed by culturing an explant comprising the foliage leaf base of a plant that propagates via scaly bulbs or bulbs is isolated and cultured.

Preferably, the explant is a foliage leaf base from which the shoot apex and the foliage leaf have been removed, and the explant is cultured in the absence of plant hormones to form a domy tissue.

Preferably, the foliage leaf base is a section from the joint to a part 1-3 mm lower therefrom of a foliage leaf.

The plants that propagate by scaly bulbs or bulbs are preferably the plants of the genus *Allium*.

Said plants of the genus *Allium* are preferably

Allium sativum.

Brief Description of the Drawing

Figure 1 is a schematic cross-sectional view of the scaly bulb of *Allium sativum*.

Best Mode for Carrying Out the Invention

(a) Applicable plants

The present invention applies to plants that form scaly bulbs having foliage leaves or bulbs. Such plants include lilies, narcissuses, tulips and the like, and most preferably plants of the genus *Allium* especially *Allium sativum*.

(b) Preparation of explants

As an explant, the present invention uses the base of a foliage leaf. Methods of extracting a foliage leaf base from a scaly bulb or a bulbs involves, for example, the sterilizing scaly bulbs or bulbs that were cut into a suitable size with a bacteriocide that has no direct effect on plant cells such as sodium hypochlorite, benzalkonium chloride, ethyl alcohol etc., washing then adequately with a sterilized water, removing the stored leaves thereby to expose the foliage leaf base, and after removing the upper part of the foliage leaf and the shoot apex, excising the base of the remaining the foliage leaf into a suitable size, for example about 1-3 mm, which is subjected to culturing (Japanese Unexamined Patent Publication (Kokai) No. 6-197650). The position of the foliage leaf base in the scaly bulb of garlic is shown in Figure 1.

(c) Culture medium used

As the culture media, any medium that contains inredients that are essential for plant growth including inorganic salts, organic salts such as vitamins, carbon sources, regulating agents of plant growth, and the like can be used. In accordance with the present invention, Murashige and Skoog medium, Linsmaier and Skoog medium

and the like can be used.

(d) Culture

An explant prepared as described in (b) is implanted into the above medium, and is cultured at a temperature (10-30°C, preferably 20-26°C) suitable for plant growth at an illumination of 50-15000 lux, preferably 3000-8000 lux (9-18 hours daily, preferably 12-16 hours), which forms a domy tissue in 5-7 days. When culture is continued, this domy tissue grows into a plant. In this culture method, the domy tissue is isolated from the explant and cultured. Domy tissues formed in 5-7 days of culture are excised with a razor blade or a scalpel, and they are again implanted in the medium shown in (c). By culturing in a similar manner, the domy tissues turn green and grow into small plantlets in about a month. When culture is further continued, the plantlets become rooted.

(e) Cultivation

The rooted plantlets obtained in the above culture (d) are implanted into a polypot containing a suitable potting compost and grown to produce seedlings. The grown plantlets are implanted into a larger pot or into the filed for cultivation.

(f) Virus testing

The plant obtained in the main cultivation is subjected to virus testing to insure that the plant is virus-free. The virus testing can be performed by a method established for each plant. For *Allium* plants, for example, there is a testing method that uses anti-virus antibody or a testing method for confirming the presence or absence of a viral gene by the PCR method (PHYTOPATHOLOGY, Vol. 86, No. 3, 253-259 (1996)).

Example

This example shows a culture method for generating virus-free plants using *Allium sativum* L. among plants of the genus *Allium*.

As a material for culturing, Fukuchi white sp. and *Allium sativum* L. native to Hokkaido were used. The strains of Fukuchi white sp. used were infected with one virus, leek yellow stripe virus (LYSV), or two viruses, leek yellow stripe virus (LYSV) and onion yellow dwarf virus (OYDV), and the strains of the species native to Hokkaido were infected with four viruses, garlic viruses (GarVs), leek yellow stripe virus (LYSV), onion yellow dwarf virus (OYDV), and garlic latent virus (GLV).

1. Sterilization of the material

Scaly bulbs of a garlic were decomposed into cloves and the outer coat was removed. Then after washing with benzalkonium chloride and water, the base of the clove was cut into sections of a 1 cm cube. The sections were immersed in 70% ethanol for 5 minutes, followed by washing with sterilized water.

2. Preparation of explants

After sterilization of the material, the remaining storage leaf portion was removed to expose the foliage leaf. The upper part of the foliage leaf was cut off so that the height of the foliage leaf was about 0.5 cm. After vertically splitting into four parts, the remaining lower part of the foliage leaf was peeled off, and the shoot apex was removed. The remaining base was cut into a section of about 2 mm in thickness to prepare an explant.

3. Medium

The Linsmaier and Skoog medium (hereinafter referred to as the LS medium) to which no plant hormones were added was used for culture.

4. Culturing

The prepared explant was implanted onto the LS medium, and subjected to a stationary culture at 25°C under illumination for 16 hours per day.

5. The formation of domy tissues and isolated culture

One week after the start of culturing, domy

tissues of about 0.5 mm in diameter were formed on some of the explants. Twenty to thirty domy tissues were formed per scale. The domy tissues were cut out with a scalpel, and implanted to the LS medium to which no hormones were added, and then subjected to a stationary culture at 25°C under illumination for 16 hours per day. The domy tissues turned green and grew into plants in 2-4 weeks. About 100% of the isolated domy tissues grew in this manner. The plants were implanted to a new culture medium, and on continued culture the plants became rooted. The rooted plants were implanted to an earth-filled polypot and cultivated.

6. Virus testing

The virus testing was performed using as test specimen the leaf blade of garlic obtained by isolated culture of the domy tissue. The virus testing was performed by RT-PCR using part of the viral gene sequence as the primer.

RNA was extracted from the leaf blade, from which cDNA was synthesized using a cDNA synthesis kit. With the cDNA as the template, PCR was performed using primers for viral detection. The primers had been designed based on the base sequence of the gene of a virus that infects garlic so as to obtain amplification products specific to the virus. Specific primers were designed for each of the four viruses, garlic viruses (GarVs), leek yellow stripe virus (LYSV), onion yellow dwarf virus (OYDV), and garlic latent virus (GLV), and a PCR reaction was performed. After the PCR reaction, the presence of specific amplification products was investigated by agarose gel electrophoresis to determine the presence of the virus.

As a result, in any of the Fukuchi white species and the Hokkaido native species, no amplification products that indicate the presence of the virus were confirmed from the plants that were obtained by the isolated culture of the domy tissues. Amplification products that

indicate the presence of the virus were confirmed in the plants obtained by a continued culture without isolating the material garlic and the domy tissues. Thus, it was confirmed that virus-free garlic can be obtained by isolated culture of the domy tissue formed in the culture of the foliage leaf base.

The primers used for viral detection were as follows:

Table 1

Primer sequence and the region to be amplified

Virus	Primer sequence			Region to be amplified
GarV-A, B, C, D (common)	N-RT1 :	5'-CCTGCTAAGCTATATGCTGA-3'	(Seq. ID. No.: 1)	3'-terminal ORF
	N-RT2 :	5'-GTAAGTTTAGCGATATCAAC-3'	(Seq. ID. No.: 2)	3' non-cording
LYSV	7-RT3 :	5'-AAGAGTCAACACTTGGTTTG-3'	(Seq. ID. No.: 3)	3' non-cording
	7-RT4 :	5'-GGTCTCAATCCTAGCTAGTC-3'	(Seq. ID. No.: 4)	3' non-cording
OYDV	OGP1 :	5'-GAAGCGCACATGCAAATGAAG-3'	(Seq. ID. No.: 5)	CP gene
garlic-type	OGM1 :	5'-CGCCACAACTAGTGGTACAC-3'	(Seq. ID. No.: 6)	3' non-cording
GLV/SLV	SLP1 :	5'-TATGCTCGAGCTCGTAGAGC-3'	(Seq. ID. No.: 7)	3'-terminal ORF
	HRT2 :	5'-GGGTTTCACATTGTTACACC-3'	(Seq. ID. No.: 8)	3'-terminal ORF
GCLV	GCP1 :	5'-AATGGGTGTTCTAGGAGTGC-3'	(Seq. ID. No.: 9)	3'-terminal ORF
	GCM1 :	5'-TTAAACCTTAGTCAAGCTATTC-3'	(Seq. ID. No.: 10)	3' non-cording
Alliinase	C-AL5' :	5'-TCTGGTAGTCGATTTGGGTGGGCG-3'	(Seq. ID. No.: 11)	
	NC3' -1:	5'-GCCGTAGCATTAGGATGTATG-3'	(Seq. ID. No.: 12)	

The result of virus testing is also shown below:

Table 2

Result of virus testing for each plant

Plant	Virus			
	GarVs	LYSV	OYDV	GLV
Fukuchi white species (one virus species)				
(1) Parent plant	-	+	-	-
(2) Regenerated plant without isolating the domy tissue	-	+	-	-
(3) Regenerated plant by isolating the domy tissue (a total of 11 plants)	-	-	-	-
Fukuchi white species (two virus species)				
(1) Parent plant	-	+	+	-
(2) Regenerated plant without isolating the domy tissue	-	+	+	-
(3) Regenerated plant by isolating the domy tissue (a total of 5 plants)	-	-	-	-
Hokkaido native species				
(1) Parent plant	+	+	+	+
(2) Regenerated plant without isolating the domy tissue	+	+	+	+
(3) Regenerated plant by isolating the domy tissue (a total of 6 plants)	-	-	-	-

Industrial Applicability

In accordance with the culture method of the present invention for generating virus-free plants, a larger amount of virus-free plants can be obtained as compared to the shoot apex culture method. In the conventional shoot apex culture method, it was common that there are only one or a few shoot apex per scale and one shoot apex produces only one plant, and thus it was difficult to obtain a large quantity of virus-free plants. In contrast, the method of culturing the foliage leaf base produces a plurality of plants: in the case of garlic, one scale produces 20-30 domy tissues. Therefore, a large scale propagation of virus-free plants is facilitated.

CLAIMS

1. A method of generating virus-free plants characterized in that a domy tissue formed by culturing the foliage leaf base of a plant that propagates via scaly bulbs or bulbs is isolated and cultured.

2. The method according to claim 1 wherein an explant comprising the foliage leaf base from which the shoot apex and foliage leaves have been removed is cultured in the absence of plant hormones to form a domy tissue.

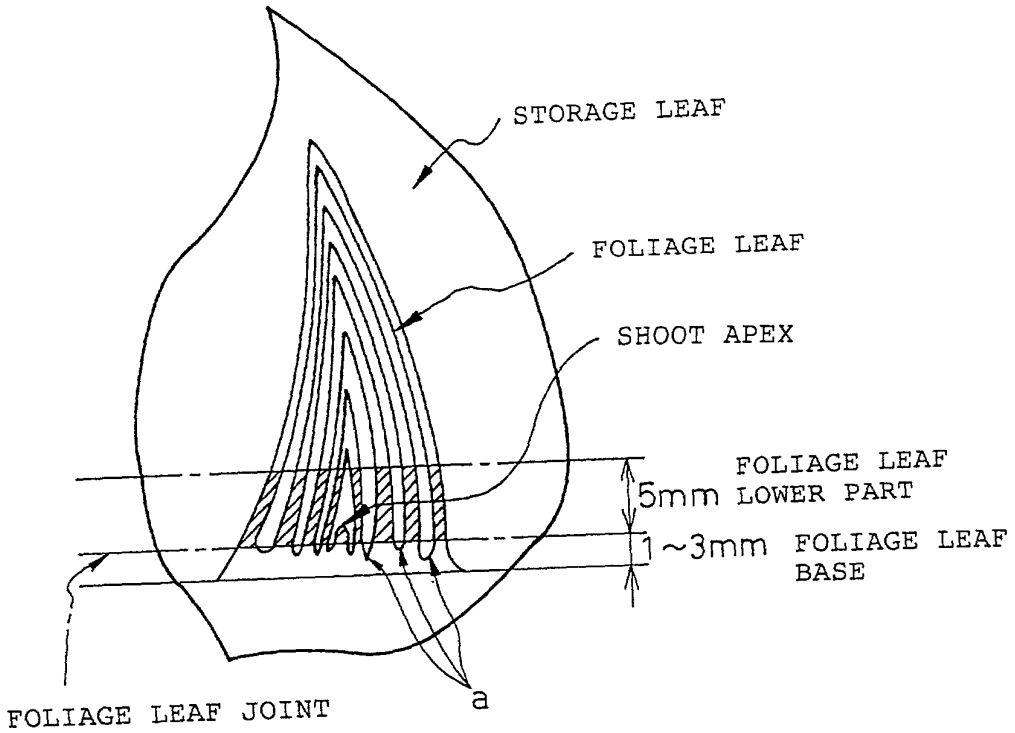
3. The method according to claim 1 or 2 wherein said foliage leaf base is a section from the joint to a part 1-3 mm lower therefrom of a foliage leaf.

4. The method according to any one of claims 1 to 3 wherein the plant that propagates with scaly bulbs or bulbs is a plant of the genus *Allium*.

5. The method according to claim 4 wherein the *Allium* plant is *Allium sativum*.

EP0612464A1

Fig.1



FOR PCT-350292001100

Declaration and Power of Attorney For Patent Application

特許出願宣言書及び委任状

Japanese Language Declaration

日本語宣誓書

私は、下欄に氏名を記載した発明者として、以下の通りに宣言します。

私の住所、郵便の宛先及び国籍は、下欄に氏名に続いて記載した通りです。

下記の名称の発明に関し、請求の範囲に記載され、特許出願している発明内容について、私が、最初にして唯一の発明者である（一人の氏名のみが下欄に記載されている場合）か、もしくは、最初にして共同の発明者である（複数の氏名が下欄に記載されている場合）と信じています。

上記発明の明細書（下記の欄でX印がついていない場合は、本書に添付）は、

□ 年 月 日に提出され、米国出願番号または特許協力条約国際番号を、
第 号とし、（該当する場合）
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As a below named inventor, I hereby declare that:

My residence, post office address and citizenship are as stated next to my name.

I believe I am the original, first and sole inventor (if only one name is listed below) or an original, first and joint inventor (if plural names are listed below) of the subject matter which is claimed and for which a patent is sought on the invention entitled:

METHOD OF GENERATING VIRUS-FREE PLANTS

the specification of which is attached hereto unless the following box is checked:

☒ was filed on June 20, 2000
as United States Application Number or
PCT International Application Number
PCT/JP00/04022 and was amended on
(if applicable)

I hereby state that I have reviewed and understand the contents of the above identified specification, including the claims, as amended by any amendment referred to above.

I acknowledge the duty to disclose information which is material to patentability as defined in Title 37, Code of Federal Regulations, Section 1.56.

I hereby claim foreign priority under Title 35, United States Code, Section 119(a) -(d) or 365(b) of any foreign application(s) for patent or inventor's certificate, or 365(a) of any PCT International application which designated at least one country other than the United Sates, listed below and have also identified below, by checking the box, any foreign application for patent or inventor's certificate, or PCT International application having a filing date before that of the application on which priority is claimed.

Japanese Language Declaration
(日本語宣誓書)

Prior foreign applications 先の外国出願			Priority claimed 優先権の主張	
11-175768(Pat.Appln.) (Number) (番 号)	Japan (Country) (国 名)	22/ June/ 1999 (Day/Month/Year Filed) (出願の年月日)	☒ Yes あり	☐ No なし
(Number) (番 号)	(Country) (国 名)	(Day/Month/Year Filed) (出願の年月日)	☐ Yes あり	☐ No なし
(Number) (番 号)	(Country) (国 名)	(Day/Month/Year Filed) (出願の年月日)	☐ Yes あり	☐ No なし

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I hereby claim the benefit under Title 35, United States Code, Section 119(e) of any United States provisional application(s) listed below.

(Application No.) (出願番号)	(Filing Date) (出願日)

(Application No.) (出願番号)	(Filing Date) (出願日)

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(Application No.) (出願番号)	(Filing Date) (出願日)	(Status: Patented, Pending, Abandoned) (現況：特許許可済み、係属中、放棄済み)
(Application No.) (出願番号)	(Filing Date) (出願日)	(Status: Patented, Pending, Abandoned) (現況：特許許可済み、係属中、放棄済み)

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(日本語宣言書)

委任状： 私は下記の発明者として、本出願に関する一切の手続きを米国特許商標局に対して遂行する弁理士または代理人として、下記の者を指名致します。（弁護士、または代理人の指名及び登録番号を明記のこと）

POWER OF ATTORNEY: As a named inventor, I hereby appoint the following attorney(s) and/or agent(s) to prosecute this application and transact all business in the Patent and Trademark Office connected therewith(list name and registration number)

Barry E. Bretschneider, Reg. No. 28,055
Raj S. Dave, Reg. No. 42,465; Richard D. Jordan, Reg. No. 33,519; Richard Lilley, Reg. No. 42,803; Paul J. Riley, Reg. No. 38,596; Kevin R. Spivak, Reg. No. 43,148

書類送付先：

Send Correspondence to:
Barry E. Bretschneider Reg. No. 28,055
Morrison & Foerster LLP
2000 Pennsylvania Avenue, N.W.
Washington, D.C. 20006-1888

直接電話連絡先：（名前及び電話番号）

Direct Telephone Calls to: (name and telephone number)
Barry E. Bretschneider
Phone: (202) 887-1545
Fax: (202) 887-0763

唯一または第一発明者名	Full name of sole or first inventor Masanori Ayabe
発明者の署名 日付	Inventor's signature Date Masanori Ayabe November 28, 2001
住所	Residence Takata-gun, Hiroshima, Japan
国籍	Citizenship Japanese
私書箱	Post Office Address C/O Wakunaga Pharmaceutical Co., Ltd. 1624, Shimokotachi, Koda-cho, Takata-gun, Hiroshima 739-1195 Japan
第二共同発明者	Full name of second joint inventor, if any Shinichiro Sumi
第二共同発明者 日付	Second inventor's signature Date November 28, 2001
住所	Residence Takata-gun, Hiroshima, Japan
国籍	Citizenship Japanese
私書箱	Post Office Address C/O Wakunaga Pharmaceutical Co., Ltd. 1624, Shimokotachi, Koda-cho, Takata-gun, Hiroshima 739-1195 Japan

（第三以降の共同発明者についても同様に記載し、署名をすること）

(Supply similar information and signature for third and subsequent joint inventors.)

10/018293
JC05 Rec'd PCT/PTO 20 DEC 2001

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